Downhill Kinetics of Biomolecular Interface Binding: Globally Connected Scenario

Jin Wang,*† Weimin Huang,* Hongyang Lu,* and Erkang Wang*
*State Key Laboratory of Electro-analytical Chemistry, Changchun Institute of Applied Chemistry, Chinese Academy of Sciences, Changchun, People's Republic of China; and †The Department of Chemistry and Department of Physics, State University of New York at Stony Brook, Stony Brook, New York

ABSTRACT We study the kinetics of the biomolecular binding process at the interface using energy landscape theory. The global kinetic connectivity case is considered for a downhill funneled energy landscape. By solving the kinetic master equation, the kinetic time for binding is obtained and shown to have a U-shape curve-dependence on the temperature. The kinetic minimum of the binding time monotonically decreases when the ratio of the underlying energy gap between native state and average non-native states versus the roughness or the fluctuations of the landscape increases. At intermediate temperatures, fluctuations measured by the higher moments of the binding time lead to non-Poissonian, non-exponential kinetics. At both high and very low temperatures, the kinetics is nearly Poissonian and exponential.

INTRODUCTION

The study of biomolecular binding is essential in the understanding of molecular recognition (Koshland, 1958; McCammon, 1998). There are two major issues related to the recognition. One is the affinity, which measures the thermodynamic stability of native binding state. The other is the specificity, which measures how a particular sequence or set of interactions is best suited for binding discriminatively from others (Wlodawer and Erickson, 1993; Clackson and Wells, 1995). In practice, rational drug design needs accurate contributions of both structure and energy to reach affinity and specificity (Cherfils and Janin, 1993; Oshiro et al., 1995). The recent progress in combinatorial chemistry opens up a new way to the drug design industry (Gallop et al., 1994; Gordon et al., 1994). By trying different possible ligand sequences, a specific ligand-receptor complex with good binding property can be picked out much like the natural evolution selection process for specificity.

Clearly quantitative analysis of binding is needed to characterize the ensemble of sequences. Although the current microscopic description of binding in terms of molecular interactions cannot guarantee a reliable answer, the statistical phenomenological description which has been successfully applied in physics of spin glasses (Mezard et al., 1987) and protein folding (Wolynes et al., 1995; Gutin et al., 1995; Dill et al., 1995) can be used to mimic the ensemble of sequences of ligand binding to a receptor or an ensemble of different kinds of interactions due to the sequence heterogeneity.

We pick the ligand-receptor complex as our model system for studying biomolecular binding. Due to the different types of interactions among amino acid residues between ligands and receptors (20 different kinds of amino acids), the interaction strengths can be approximated as Gaussian-distributed. The resulting statistical energy landscape has roughness or fluctuations characterized by the width of the distribution. However, because of the vast number of degrees of freedom, if one has to randomly search through all the state space, it takes cosmological time to reach the native binding state. In reality, binding happens in milliseconds to seconds. This is the so-called Levinthal paradox (Levinthal, 1969). To resolve this issue, it is natural to assume that there is an energy bias toward the native binding state. In this way, one expects the binding process to complete, in real time, toward the native state. In other words, there exists a funneled shape landscape leading toward native binding state while superimposed on this funnel is the roughness or fluctuations of the energy landscape (Rejto and Verkhivker, 1996; Tsai et al., 1999, 1998; Tovchigrechko and Vakser, 2001; Shoemaker et al., 2000; Papoian and Wolynes, 2003; Wang and Verkhivker, 2003; Levy et al., 2004). There are in general multiple paths leading toward the native binding states at initial stage of binding, the discrete paths emerge at later stage of binding when the energy landscape becomes rough and there exists possible local minima that traps the system similar to the protein-folding problem (Wang et al., 1996).

The thermodynamics of the binding energy landscape has been discussed in detail recently (Shoemaker et al., 2000; Papoian and Wolynes, 2003; Wang and Verkhivker, 2003). There exists, in general, several phases—the non-native unbinding phase, the native binding phase, and the local trapping phase. To avoid the local trapping or glass phase reaching the native binding phase, the maximization of the ratio of the transition temperature to the native binding state versus the local trapping temperature is required. It was also shown that this is equivalent to the requirement of the maximization of the ratio of the energy gap between the native binding state and the average of non-native states versus fluctuations or spread of the non-native states. In other

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Address reprint requests to Jin Wang, E-mail: jinwang@sprynet.com, or to Erkang Wang, E-mail: ekwang@ciac.jl.cn.

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words, the binding phase should be far from the glass phase to avoid falling into the traps of local minimum. This is also the optimal criterion for specificity. Only those sequences of biomolecules (ligands, in this example) or a set of interactions satisfying this criterion can bind discriminately against others.

Whereas the simple model of thermodynamics of the binding energy landscape has been studied, the kinetics of the binding process has not yet been explored much theoretically. This is the purpose of the current study.

First, we will choose an order parameter or reaction coordinate that captures the physics of the kinetic process. In accordance to the thermodynamic model we studied, this order parameter is Q, where Q is the fraction of native spatial contacts between the ligand and the receptor of the biomolecular binding complex at the binding interface ($Q = 0$ when the system is in the completely unbinding states, and $Q = 1$ when the system is in the native binding state). The hydrophobic and electrostatic short-range interactions play important roles in the energetics between ligand and receptor, and can be approximately represented using the order parameter Q.

To study the kinetics process from the non-native unbinding states to the native binding state, we have to specify how the configurational states are connected (Leopold et al., 1992). When the corresponding underlying free-energy landscape is a downhill funnel in Q so that the binding process at the interface is activationless or when the underlying thermodynamic free-energy driving force is large, the states are more likely to be globally connected in Q. In this situation, any state is likely to be connected to the other states, which can have quite different values of Q, and the kinetics-moves from one Q to another value of Q can be fast. We believe that this situation is suitable for the description of the many ligand-receptor binding complexes at the interface. This situation is clearly shown in Fig. 1.

In principle, both local and global connectivity exist to some extent between conformational states (Fig. 1). One needs to take them into account carefully (Wang, 2003). Of course, in the multidimensional configurational space, states are locally connected. The global connectivity only appears in the reduced representation with a single order parameter Q describing the binding process.

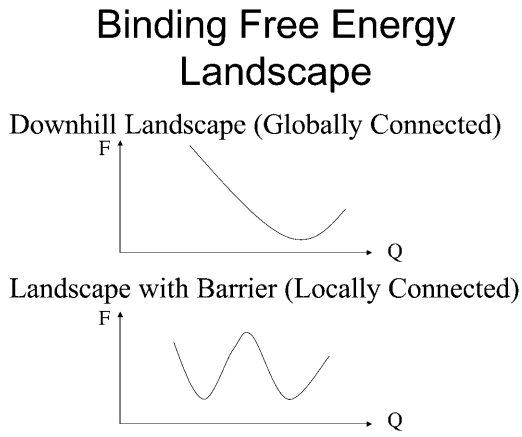

FIGURE 1 The downhill free-energy landscape in Q and free-energy landscape with activation barrier in Q.

MATERIALS AND METHODS

We start the discussions on the energy landscape of binding using the random energy model with the bias toward the native state. A similar problem was studied in the context of protein folding (Plotkin et al., 1996, 1997; Wang et al., 1996; Saven et al., 1994). For simplicity, we will assume that the transition state between any two energetic levels has the same energy for all such kinetically connected pairs. For many physical situations, large structural and conformational changes may be required in exploring the relevant underlying binding energy landscape. Topological and steric constraints may require a specific binding conformation state to completely unravel before a new conformational state may be reached. In such situations, many states are likely to share a common activation energy—the unraveling one. Let us begin with the kinetic master equation that describes the probability between the states of this model as

$$dP_i/dt = \sum_j k_{ij}P_j(t) - \sum_j k_{ji}P_i(t), \tag{1}$$

where $P_i(t)$ is the probability of being in state i at time t, and k_{ij} is the rate of going from state j to state i. We will specify the kinetic jumping probability according to Metropolis dynamics and assume all states have the same activation energy $\bar{E}$. Then the kinetic rate for jumping from the state j to i is given by $k_{ij} = \gamma_j k_j \exp[-\beta(\bar{E} - E_j)] = \gamma_j k_j \exp[\beta E_j]$, where $\kappa_j = k_j \exp[-\beta\bar{E}]$. The value γ_j is the probability that site j is connected to the geometrical and topological constraints. The connectivity in this model only depends on the originating site.

The order parameter Q ($0 <= Q <= 1$), the fraction of native spatial contacts at the binding interface, is a measure of how similar a particular configuration is to the native state; $Q = 1$ when the ligand-receptor complex is in its native state. In the Laplace space s, by solving the master equation and summing over the states in each stratum Q, we can obtain the probability of being in a state with order parameter Q at a particular value of s, $\bar{P}(Q, s)$, as

$$\bar{P}(Q, s) = \frac{Z(Q, s)}{sZ(s)}, \tag{2}$$

where

$$Z(Q, s) = \sum_{j=1}^{\Omega(Q)'} \frac{1}{(s + \kappa_j W_j exp[\beta E_j])}, \tag{3}$$

and where $W_i = \gamma_i \Omega$ is the average number of the states that are accessible from any given state (Ω is the total number of states in the system). The prime symbol ($'$) means that this sum is only through those states with order parameter Q, of which there are $\Omega(Q)$ such states. Note that we can also write $Z(s)$ as $Z(s) = \sum_{NQ=0}^{N} Z(Q, s)$, where N is the total number of native contacts. The expression of $Z(Q, s)$ and $Z(s = 0)$ reminds us of the equilibrium partition function. Here we can call $Z(Q, s)$ the frequency-dependent state partition function.

From the thermodynamic study of the binding landscape (Shoemaker et al., 2000; Papoian and Wolynes, 2003; Wang and Verkhivker, 2003), we know that the glass transition temperature that traps the binding complex is defined as

$$T_g = 1/\beta_g(0) = \sqrt{\frac{\Delta E^2(Q=0)}{2S(Q=0)}}, \quad (4)$$

where $S(Q=0)$ is the total configurational entropy of the binding complex. $\Delta E(Q)$ is roughness or variance of the energy landscape, which is defined below. The number of states with the order parameter Q and energy E in the interval E and $E + dE$ is

$$\Omega(Q,E)dE = \Omega(Q)g(Q,E)dE, \quad (5)$$

where the Gaussian distribution of a binding energy landscape biasing toward the native state is given by

$$g(E,Q) = \frac{dE}{\sqrt{2\pi\Delta E^2}}\exp[-(E-\bar{E}(Q))^2/(2\Delta E^2)], \quad (6)$$

where $\bar{E}(Q) = -|\delta\epsilon|\,QN$ ($\delta\epsilon$ is the native energy strength per contact) and $\Delta E^2(Q) = N\Delta\epsilon^2(1-Q^2)$ for underlying two-body interactions. ($\Delta\epsilon$ is the energy variance strength per contact). Note that $\int_{-\infty}^{\infty} = 1$ and $\Omega(Q) = \exp[S(Q)]$ (Plotkin et al., 1996, 1997).The free energy of the model is thus given by Wang and Verkhivker (2003) as

$$F(Q,T) = \bar{E}(Q) - \Delta E^2(Q)/2T - TS(Q) \quad (7)$$

for $T > T_g$, and as

$$F(Q,T) = \bar{E}(Q) - \Delta E^2(Q)/T \quad (8)$$

for $T <= T_g$.

Here the configurational entropy is $S(Q) = k_b \mathrm{Log}\Omega(Q)$. Above T_g, we can treat the energy distribution continuously, and $Z(Q,s)$ becomes

$$Z(Q,s) = \int_{-\infty}^{\infty} dE\Omega(Q,E)/(s + \kappa W\exp[\beta E]) = \Omega(Q)\int_{-\infty}^{\infty} dE g(E,Q)/(s+\kappa W\exp[\beta E]). \quad (9)$$

The above expression can be calculated numerically. We now find s_b, the binding rate, or inverse binding time, $1/\tau_b$. We will define a biomolecule as being in the native binding state if its value of Q is larger than some critical value Q_b. We will define s_b as the value of s at which the probabilities of the biomolecules binding or unbinding are equal, $\sum_{NQ=0}^{NQ_b}\bar{P}(Q,s_b) = \sum_{NQ=NQ_b+1}^{N}\bar{P}(Q,s_b)$. We see that s_b also satisfies $\sum_{NQ=0}^{NQ_b} Z(Q,s_b) = \sum_{NQ=NQ_b+1}^{N} Z(Q,s_b)$. And so by finding the root of this equation, we may obtain an estimate of the rate of binding. In the two-state model of the binding process, the above equation reduces to $Z(0, s_b) = Z(1, s_b)$, where $Q = 0$ and $Q = 1$ correspond to the unbinding and binding states, respectively.

It is important to mention that from the thermodynamic study of the binding energy landscape, we have the relation (Goldstein et al., 1992; Abkevich et al., 1994; Klimov and Thirumalai, 1998)

$$T_b/T_g = \Lambda + \sqrt{\Lambda - 1}, \quad (10)$$

where $\Lambda = \sqrt{\delta E/S\Delta E}$. Here δE is the gap between the native binding state and the average of non-native binding states. ΔE is the fluctuation or variance of the binding energy landscape. S is the total configuration entropy. The thermodynamic criterion is the optimization of T_b/T_g to guarantee the binding specificity in discriminating the native binding state from the non-native binding states. Since Λ monotonically depends on T_f/T_g, it is a good measure of specificity. We thus term Λ the specificity ratio.

RESULTS

In Fig. 2, we show the long-time binding probability (equilibrium probability) at different values of temperature with respect to the specificity measure—the gap/roughness ratio of the energy landscape. The equilibrium probability shows a relatively sharper transition at lower temperatures with higher probability as the specificity (the gap/roughness) ratio increases to a value >1. It reflects the fact that the native binding state is preferred at lower temperature and specificity increases the native stability. It helps to explain why the native binding state is so favored under this situation. When the gap/roughness ratio decreases to a value <1, non-native unbinding states are preferred.

In Fig. 3, the long-time binding probability versus inverse specificity (roughness/gap) ratio is shown. We notice that the equilibrium probability is much more sensitive to the roughness than to the gap. For each temperature, the equilibrium probability decreases with increasing the roughness. The transition from a high probability to a low probability of binding is sharper at higher temperature, reflecting the fact that non-native unbinding states are preferred at high temperatures.

In Fig. 4, we show the long-time binding probability of binding at different temperatures versus the binding affinity (the free-energy difference between the native and non-native binding states). It is the analog of the titration curve for thermodynamic binding. It shows similar behavior to that depicted in Fig. 1, with respect to the specificity measure. The equilibrium is shifted from the non-native binding states toward the native binding state as the affinity increases. This transition of equilibrium becomes sharper as the temperature decreases. This shows that the affinity and specificity can be correlated.

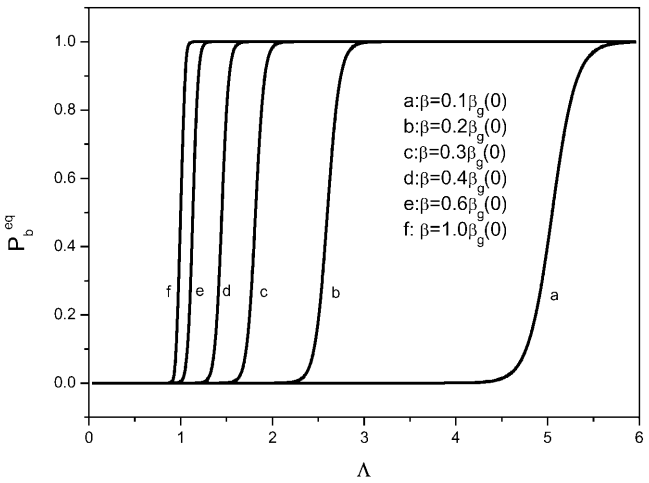

FIGURE 2 Long-time equilibrium probability of binding versus specificity ratio of different temperatures.

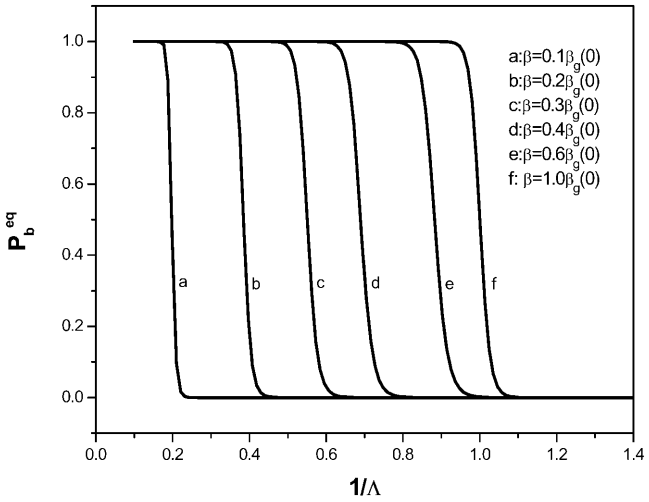

FIGURE 3 Long-time equilibrium probability of binding versus inverse specificity ratio of different temperatures.

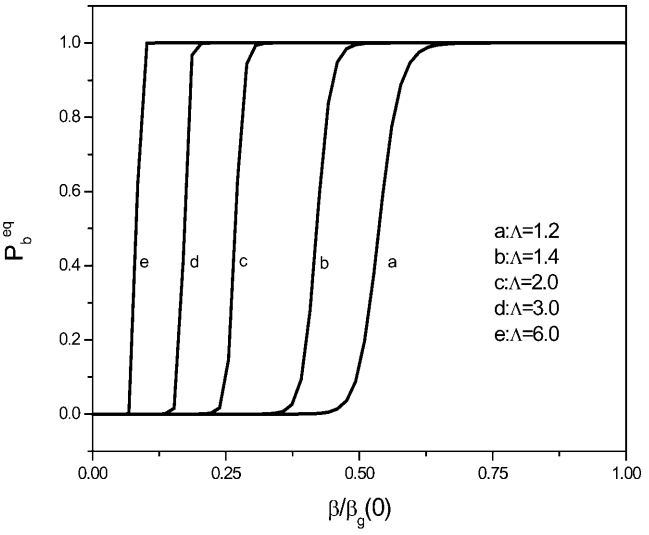

FIGURE 5 Long-time equilibrium probability of binding versus inverse temperature of different specificity ratios.

In Fig. 5, we show the long-time binding probability versus temperature for different specificity ratios. There exists a transition from the non-native states to a native state of binding when the specificity ratio is >1. This transition becomes sharper as the specificity ratio increases. This is due to the fact that high specificity discriminates the other non-native states from the native state, leading to better and sharper separation.

The plot of the binding time $\tau_b = 1/s_b$ versus temperature is shown in Fig. 6. The first feature we notice is that the binding time has a U-shape curve-dependence on temperature. This can be easily understood. As the temperature increases, the binding complex becomes less and less stable, and the non-native unbinding states are more and more preferred. Therefore the time it takes to reach the native state is longer. On the other hand, as the temperature decreases, the time it takes to reach the native binding state is also longer. This is because it is more and more likely that the complex will be trapped into some low-lying energy states (local minimum).

The optimal binding time is at the temperature of kinetic minimum. In terms of kinetics, when the ratio T_b/T_g or specificity ratio Λ is maximized, the binding time is minimized. So the high specificity ratio guarantees the thermodynamic stability as well as the kinetic accessibility. This feature can be seen from Fig. 7, where we plot τ_{min} as a function of Λ.

We can also study the fluctuations of the kinetic times. This can be realized by the fact that $s = 1/\tau$ and we know the distribution of $P(Q, \tau)$ is approximately related to the distribution of $\bar{P}(Q, s)$, which we know as shown above by the identity $\bar{P}(Q, s)ds = P(Q, \tau)d\tau$ where we obtain

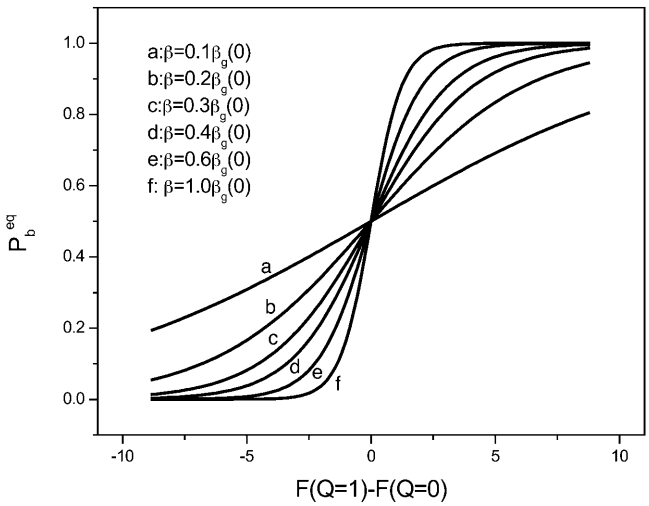

FIGURE 4 Long-time equilibrium probability of binding versus affinity of different temperatures.

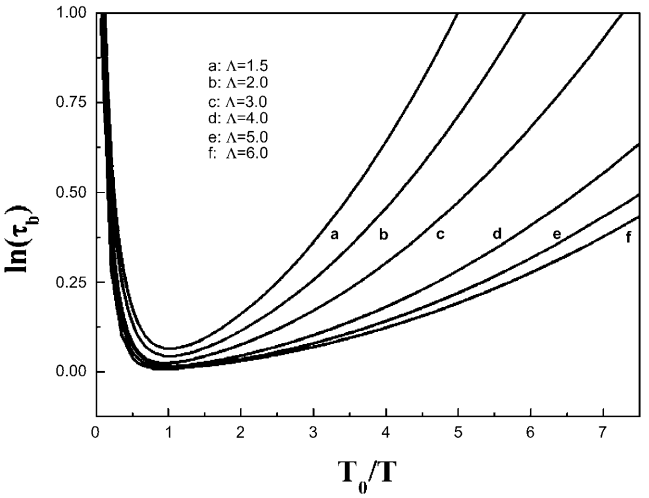

FIGURE 6 The mean first passage time versus inverse temperature of different specificity ratios.

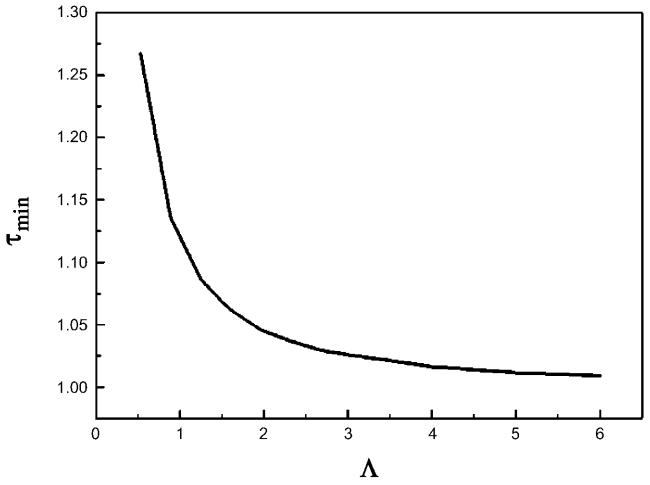

FIGURE 7 The mean first passage time at kinetic minimum temperature versus specificity ratio.

$P(Q,\tau) = \bar{P}(Q, 1/\tau)ds/d\tau$. We plot the ratio of the second moment to the square of the average of the binding time $\langle\tau^2\rangle/2!\langle\tau\rangle^2$ with respect to temperature in Fig. 8. We see at high temperatures that the ratio drops close to 1, indicating a nearly-Poissonian exponential kinetics. For a Poissonian process, the moments satisfying the condition are $\langle I^n\rangle = n!\ \langle I\rangle^n$; a Poissonian distribution implies an exponential kinetic process (Wang, 2003). On the other hand, when the temperature drops, the ratio increases and becomes significantly >1. The kinetics starts to deviate from a Poissonian process. In fact, the kinetics is quite non-exponential. At very low temperature, the ratio drops close to 1, indicating a nearly-Poissonian exponential kinetics again.

DISCUSSIONS

The specificity criterion mentioned has its limit. In nature, the binding should indeed be stable, specific, and kinetic-accessible. In addition, the biomolecular binding complex should have a certain flexibility for the functional purpose. This implies that the local transition rates from native to non-native states cannot be too small. The finite value of the local transition rate can serve as a constraint or upper bound to the specificity ratio (since the binding time decreases as the specificity ratio increases, as shown in Fig. 6). The actual optimization is the balance among the stability, specificity, kinetic accessibility, and flexibility for function.

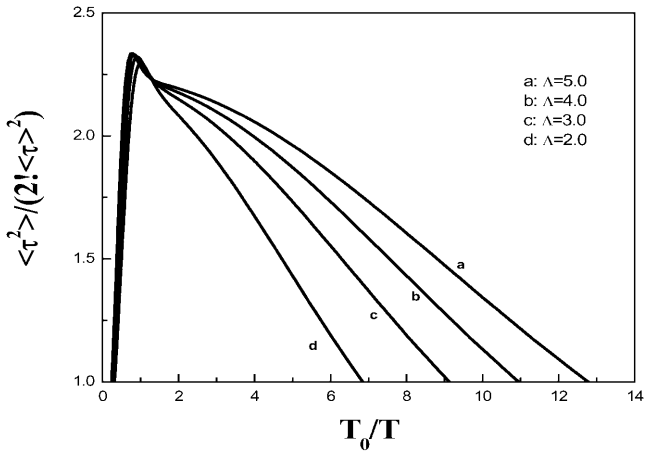

FIGURE 8 Ratio of second moment and square of the first moment versus inverse temperature of different specificity ratios.

There is a simple physical explanation of the temperature-varying fluctuations of the binding kinetics—namely, the Poisson(exponential)-non-exponential-Poisson(exponential) transition at various temperature regimes. At high temperatures, only the global features of the underlying landscape are revealed. There are, in general, multiple parallel pathways leading toward the native binding state. Each path gives a similar contribution to the kinetics. The resulting kinetics is thus a single exponential and the process is nearly Poissonian. At lower temperature below T_0, more and more local traps become important. In general, the discrete kinetic paths emerge and are distinct from each other. The kinetics is thus often non-exponential. At very low temperature, only very few states on the landscape are kinetically accessible. The kinetics therefore is dominated with the energy barrier in the deepest valley into which the system is trapped, resulting in a nearly-exponential process and Poissonian statistics again.

There have been many binding experiments on the temperature dependence of the kinetics to explore the relationships among the structures, functions, and dynamics of the underlying binding energy landscapes (Frauenfelder et al., 1991 and references therein). As a concrete example in connection with the experiments, we look at the binding of a small ligand, carbon monoxide (CO), with a protein myoglobin, (Mb). This is a model system, which has been thoroughly investigated experimentally (Frauenfelder et al., 1991). In particular, by flash photolysis and kinetic hole burning, the rebinding of CO to Mb has been studied over wide ranges in time (100 ns to 1000 s) and temperature (10–320 K). At low temperatures ($T < 240$ K), non-exponential heme pocket binding kinetics has been observed as seen clearly from Fig. 1 of Steinbach et al. (1991). The degree of non-exponential kinetics characterized by the stretched exponential ($\exp[-(kT)^{\beta}]$) coefficient β seem to increase as the temperature decreases (Post et al., 1993). Furthermore, non-Arrhenius behavior of the kinetic-rate dependence on the temperature has been observed (Steinbach, et al., 1991). The rate dependence on the temperature has been fitted well with Ferry's law $k(T) = A\exp[-(E/RT)^2]$ or the kinetic relaxation time $\tau(T) = 1/A\exp[(E/RT)^2]$ in these low temperature ranges (Ferry et al., 1953), as shown in Eq. 18 and Figs. 10 and 11 of Steinbach et al. (1991). A kinetic anomaly is observed starting from 180 K to 220 K where the binding process is slower when the temperature increases, as shown in Fig. 11 of Steinbach et al. (1991). At high temperatures, equilibrium fluctuations set in; the kinetic process involves both the heme

pocket binding and escaping to (from) the solvents, and the long-time exponential pocket-binding kinetics is observed due to the rapid fluctuations or relaxations of the protein. This is also seen from Fig. 1 of Steinbach et al. (1991).

The distribution of the barriers and the relaxations or fluctuations of the protein have been used to interpret the low-temperature kinetic behavior and describe some features seen at high temperatures (Agmon and Hopfield, 1983; Steinbach et al., 1991; Panchenko et al., 1995). Our binding theory in this article, by explicitly considering the conformational changes and fluctuations at the interface, can explain reasonably well the qualitative trends of the kinetic data observed. In Fig. 6, the kinetic binding time has a minimum (or rollover point) at T_0. If T_0 is $\sim$180 K, then this figure shows the kinetic anomaly in the CO-Mb globin experiments (Steinbach et al., 1991). The kinetic anomaly of $T > T_0$ (slower kinetics when increasing the temperature) can then be explained as due to the onset of the distribution of conformational changes as the temperature increases, and the corresponding effective increase of the free-energy barrier between the more stable unbinding configurations and the less stable binding configuration as the temperature increases. The kinetic time (on logarithmic scale) plot in Fig. 6 has a U-shape and can be fitted with linear plus quadratic function. Below T_0 (corresponding to the temperatures on the right-hand side of the $T_0/T > 1$ axis), the corresponding temperature-dependence of the kinetic time (or rate) is clearly not Arrhenius-type and can be fitted to the same form as Ferry's law, mentioned in the above paragraph (Ferry et al., 1953, Steinbach et al., 1991). The explanation of this non-Arrhenius temperature-dependence of the binding time or rate comes from the motions among the distribution of the free-energy conformation valleys or traps at low temperatures. In Fig. 8, the fluctuations of the kinetic time are shown and the large fluctuations are observed near and below T_0 ($\langle\tau^2\rangle/2!\langle\tau\rangle^2 > 1$). This implies a non-Poissonian process and therefore a non-exponential kinetics (Wang, 2003) at low temperatures, consistent with the CO-Mb binding experiments (Steinbach et al., 1991) as mentioned in the above paragraph. This non-exponential kinetics is due to the onset of the distribution of the free-energy traps at low temperatures. From Fig. 8, at temperatures significantly higher than T_0, the fluctuations in kinetics are less, and the kinetics more and more exponential. This is due to the fact that, at high temperatures where fluctuations and relaxations are fast, multiple kinetic paths play a role and will lead to exponential kinetics. This is consistent with the high-temperature behavior of the CO-Mb binding experiments (Steinbach et al., 1991). From Fig. 8, the degree of the non-Poissonian nature of the process (or non-exponential kinetics) measured by $\langle \tau^2 \rangle /2! \langle \tau \rangle^2$ decreases as the temperature decreases below T_0. This is consistent with the experiments mentioned in the above paragraph where the degree of non-exponential kinetics decreases (becomes more exponential) as the temperature decreases (Post et al., 1993). This reflects the fact that as the temperature decreases, more and more conformational states are frozen, and fewer and fewer states are kinetically accessible. The kinetics, therefore, is dominated with fewer energy barriers. This leads to a lesser degree of non-exponential process. The binding model in this article predicts that, as the temperature drops to very low, the kinetic process should be dominated with the deepest valley into which the system is trapped, resulting in a nearly-exponential process and Poissonian statistics again. In the experiments (Steinbach et al., 1991), this kinetic phenomenon is not seen. This is probably due to the fact that the temperature probed is not low enough or that the experiments are carried out at the bulk level so the inhomogeneous distribution of the protein samples can cause the non-exponential kinetics at very low temperatures. This motivates the next generation of the kinetic-binding experiments to be carried out in single molecules and with wide temperature ranges to explore the detail structures of the underlying binding energy landscapes.

The high-to-low temperature, nearly-single-exponential to non-exponential transitions in kinetics has also been observed experimentally for folding of proteins (Nguyen et al., 2003). It is not surprising that the folding exhibits similar behavior since the underlying driving hydrophobic force causing the large conformational change for folding and binding are similar. The only major difference between folding and binding is the connectivity. Folding can thus be seen as self-binding. The non-exponential behavior is also observed in single-molecule enzymatic dynamics experiments (Lu et al., 1998; Yang and Xie, 2002a,b; Yang et al., 2003; Xie, 2002). This can be due to the distributions of the conformational fluctuations of the underlying energy landscape. Wide-temperature-range explorations are needed, and are yet to be done.

The investigation of the opposite case—diffusion limit where the local connectivity in order parameter Q space is important, and the free-energy barrier is important—will be given in another publication (J. Wang, unpublished).

The fluctuations and distribution of the kinetic time is often needed for uncovering the nature of the underlying energy landscape of the biomolecules, single molecules experiments (Moerner, 1996; Lu et al., 1998; Zhuang et al., 2000, 2002; Deniz et al., 2000; Jia et al., 1999), and the interrelationships among structure, function, and dynamics (Frauenfelder et al., 1979, 1991; Lee et al., 2003, 2003; Zhou et al., 2003). In single-molecule experiments, it is now possible to measure not only the mean but also the fluctuations and moments as well as the distribution of dynamical times (Zhuang et al., 2000, 2002; Deniz et al., 2000; Jia et al., 1999; Schuler et al., 2002; Lipman et al., 2003). The information on the kinetic-time distribution can be accessible through the analysis of trajectories (for example, single-molecule fluorescence) from the experiments and from the simulations (Zhou et al., 2003). So the theory/simulations and experiments can be directly qualitatively and quantitatively compared with each other. This topic is very interesting and worth further study.

The binding kinetics of short-range interactions is considered here. It would be ideal to combine this with the long-range electrostatic guiding interactions for the full-range study.

In this article, we only study the kinetics involving the conformational changes near the binding interface. It is not uncommon that binding involves large conformational changes, sometimes unfolding (Koshland, 1958; Shoemaker et al., 2000; Papoian and Wolynes, 2003). Further dynamic studies are needed to take the binding and folding coupling into account (Papoian and Wolynes, 2003).

CONCLUSIONS

We have in this report considered the kinetics of binding of biomolecules at the interface with global connectivity. We believe this is a good approximation for many ligand-receptor binding complexes where kinetics is relatively fast and the process is nearly activationless. The global connectivity is also an appropriate description when the thermodynamic driving force is large. The kinetic time constant for binding has a U-shape curve-dependence on temperature. It increases at high temperature as temperature increases due to the instability of the native binding state, whereas at low temperature the time constant of binding increases as temperature decreases due to the trapping at local minimum. This rollover behavior is observed in many kinetic experiments involving large conformational changes (see Kaya and Chan, 2000, 2002, for the summary and the references therein). The fluctuations of the binding-energy landscape also lead to temperature-varying kinetics.

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